CONCLUSION

In view of the above amendments and remarks, it is believed that all claims are in condition for allowance, and it is respectfully requested that the application be passed to issue. If the Examiner feels that a telephone conference would expedite prosecution of this case, the Examiner is invited to call the undersigned at (781) 861-6240.

Respectfully submitted,

HAMILTON, BROOK, SMITH & REYNOLDS, P.C.

By Lisa M. Treannie

Lisa M. Treannie
Registration No. 41,368
Telephone (781) 861-6240
Facsimile (781) 861-9540

Lexington, Massachusetts 02421-4799
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MARKED UP VERSION OF AMENDMENTS

Specification Amendments Under 37 C.F.R. § 1.121(b)(1)(iii)

Replace the paragraph at page 10, lines 13 through 20, with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figures 1A-1O are [is a] fluorescence activated cell scanning (FACS) histogram profiles illustrating that mAbs 1D9 and 8G2 stain CCR2 transfectants but not CCR5 or CCR1 transfectants. L1/2 (also referred to herein as L1.2) murine pre-B lymphoma host cells were transfected with CCR2, CCR5 and CCR1 as indicated, and stained with antibodies with different receptor specificities. Staining was analyzed by flow cytometry.

Replace the paragraph at page 10, line 21, through page 11, line 2, with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figures 2A-2L are [is a] FACS dot plots showing expression of CCR2 on most monocytes, a subpopulation of lymphocytes and a small subset of granulocytes. Whole blood cells were stained with one of three anti-CCR2 mAbs (5A11, generated using a peptide consisting of the first 32 amino acids of the CCR2 amino-terminus as an immunogen, and 1D9 and 8G2 generated as described herein using CCR2b L1/2 cell transfectants as the immunogen). Staining was analyzed by flow cytometry, and the lymphocyte, granulocyte and monocyte populations were gated using the forward and side light scatter. The X-axis represents forward light scatter (a measure of cell size), and the Y-axis fluorescence intensity of staining for CCR2. The level of negative control staining is indicated by a line.

Replace the paragraph at page 11, lines 3 through 12, with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

Figures 3A-3I are [is a] FACS dot plots showing that mAb 1D9 stains an IgE positive population in peripheral blood (basophils) using two-color staining for IgE and CCR2. Whole blood cells were first stained with either a negative control antibody (anti-Flag), anti-CCR2 antibody 1D9, or an anti-CXCR1 antibody, as indicated, and detected by an anti-mouse-FITC conjugate. A second staining was done using either PBS or a biotinylated antibody specific for IgE or CD16, as indicated, and detected with a streptavidin-phycoerythrin. Staining was analyzed by flow cytometry.

Replace the paragraph at page 60, lines 2 through 18, with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

For the staining of cultured transfectant cell lines 0.5 x 10^6 cells in 50 µl were resuspended in PBS + 1% FCS in a 96 well polystyrene V-bottom plate. 50 µl of primary antibody supernatants or HT medium (negative control) were added, and the samples were incubated at 4°C for 30 min. 100 µl of PBS were added and the cells were pelleted by centrifugation and washed once with PBS. The pellet was resuspended in 100 µl PBS + 1% FCS containing FITC-conjugated goat anti-mouse IgG antibody (a 1:250 dilution) and incubated for thirty minutes at 4°C in the dark. The cells were washed twice with PBS, resuspended in PBS, and analyzed by flow cytrometry with a FacScan cytometer using the CellQuest software (Becton-Dickenson) Cells were fixed with PBS/1% formaldehyde if they were not to be analyzed the same day. Monoclonal antibodies 1D9 and 8G2 stain CCR2 transfectants but not CCR1 or CCR5 transfectants (Figures 1A-1O).

Replace the paragraph at page 60, line 20, through page 61, line 13, with the below paragraph marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph.

100 µl whole blood was mixed with 100 µL of 1D9 antibody hybridoma supernatants or HT medium (negative control) and incubated at 4°C for 30 min. After one wash with PBS, 100 µL FITC-conjugated goat anti-mouse IgG antibody (a 1:250 dilution) was added to each sample and incubated for 30 min. at 4°C in the dark. Samples were then washed once with PBS if a second color staining is to be done, otherwise washed twice more in PBS. For two color staining 5 µl of mouse serum was added to the cell pellets after the single wash, mixed, and incubated for five minutes at 4°C in the dark. Second primary antibodies (or PBS as a negative control) were added (10 µl anti-CD16, 100 µl 1:200 dilution of anti-IgE) and incubated for thirty minutes at 4°C in the dark. Samples were then washed one time with PBS and resuspended in 100 µL streptavidin PE (1:200 PBS + 1% BSA) and incubated for fifteen minutes at 4°C in the dark. Eyrythrocytes were lysed by adding 2 ml of FACS Lysing Buffer to each sample and incubating at room temperature in the dark for fifteen minutes or until samples were clear. The cells were pelleted by centrifugation and all but 200 µl of the supernatant was aspirated. The samples were analyzed by flow cytometry on a FacScan cytometer using the CellQuest software (Becton-Dickenson). CCR2b is expressed on most monocytes, a subpopulation of lymphocytes and a subset of granulocytes (Figures 2A-2L). CCR2b is expressed on an IgE-positive population in peripheral blood (basophils)(Figures 3A-3I).